Pedobiologia **33**, 361–371 (1989)
VEB Gustav Fischer Verlag Jena

Institute of Soil Biology, Czechoslovak Academy of Sciences, České Budějovice, Czechoslovakia

The influence of Zeazin 50 on Enchytraeidae (Oligochaeta) in an apple orchard soil

JOSEF CHALUPSKÝ, jr.

With 3 figures

(Accepted: 89-04-19)

1. Introduction

Herbicides are the most frequently used pesticides in agriculture. Besides having a toxic effect on higher plants, their application can cause temporary as well as long term changes in the structure and function of soil biota. These changes arise from the direct action of herbicides on soil organisms, as well as from indirect effects, such as changes in microclimate, food resources, and in relations between the organisms. EIJSACKERS & DRIFT (1976) and EIJSACKERS & BUND (1980) updated the information on the influence of herbicides on soil fauna, and they suggested that none of the soil organisms studied remained completely unaffected. The information concerned mainly the herbicides DNOC, 2,4-D, TCA, dalapon and simazine and their influence on Lumbricidae, Nematoda, Acari and Collembola. Generally, the effect of phenoxy acids on the abundance of soil animals was mild, whereas the triazines and herbicides based on urea and phenol were distinctly inhibitory.

The influence of herbicides on enchytraeids has attracted less attention. In most studies enchytraeid abundance decreased after the application of herbicides. For example, in the study by POPOVICI *et al.* (1977) on the influence of atrazine in doses 5 kg and 8 kg per hectare, enchytraeid abundance had decreased by 66% and 73% one month after the treatment, and after 3 months abundance had declined further, about 55% and 19% compared with the control.

The aim of the present study was to apply the herbicide Zeazin 50 in various doses to determine its influence on the abundance, biomass and species composition of Enchytraeidae in the soil of an apple orchard. The study was part of a research project undertaken in 1981–1985 by the Laboratory of Soil Biology, Institute of Landscape Ecology, Czechoslovak Academy of Sciences.

2. Objectives of study, materials and methods

The influence of Zeazin 50 applied once in various doses was studied in a field experiment in 1981–1982.

The experiment was performed in a previously chemically untreated, 25 year old apple orchard near Bavorov village in South Bohemia (445 m asl, 49° 07′ N, 14° 05′ E). The soil type of the locality was brown soil, the humus form mull, and the soil *p*H 5.62. The plant association belonged to the alliance Arrhenatherion. Rainfall and mean air temperatures were 642 mm and 8.3°C in 1981, and 463 mm and 8.6°C in 1982. Average values for the period 1900–1950 were 586 mm and 7.3°C.

At the site, 5 experimental plots were arranged; plots were 5 m × 10 m and were separated by a 2 m buffer zone. The plots were treated with a water suspension of Zeazin 50 in doses of 0, 5, 10, 20 and 40 kg per hectare on 28th May, 1981. In the following text the plots are labelled by numbers which agree with the dose used. Control – 0 kg per hectare; variant 5 – 5 kg per hectare; variant 10 – 10 kg per hectare; variant 20 – 20 kg per hectare; and variant 40 – 40 kg per hectare. The manufacturer recommends 4–5 kg per hectare for depletion of understorey in fruit orchards, and 10–20 kg per hectare for depletion of unwanted vegetation in untilled land. The active ingredient in Zeazin 50 is atrazine (50% of volume). The maximum concentration in water solution is 33 $mg \cdot l^{-1}$.

In 1981 8 series of samples were collected. The first was on the day of application, an hour before treatment. The second was 1 week (3rd June) and the third was 3 weeks (17th June) after treatment. The remainder were collected at monthly intervals on 8th July, 12th August, 16th September, 13th October and 18th November. Nine monthly samplings were carried out in 1982 on 18th March, 19th April, 19th May, 21st June, 26th July, 18th August, 22nd September, 25th October and 26th November.

Individual soil samples were taken using a metal tube corer with a working area of $10\,cm^2$. The depth was 10 cm. In 1981, 10 soil cores were cut from the control and from variant 40 on each sampling occasion, whereas 5 cores were cut from variant 10 while variants 5 and 20 remained unsampled. In 1982, 10 cores were collected from all experimental plots on each sampling occasion. A stratified random procedure (ABRAHAMSEN, 1969) was used to determine sample location. The soil samples were divided immediately into 0–5 cm and 5–10 cm layers, put in plastic bags, transported to the laboratory and stored at 3 °C. Within the following week the enchytraeids were extracted in heated wet funnels (O'CONNOR, 1955). The worms were identified alive in a drop of tap water. Determination was to genus in 1981 and to species in 1982. Material was fixed in 4 % formaldehyde and later stored in 70 % ethanol.

The enchytraeid fauna was characterized by abundance, biomass, mean mass of single individuals (calculated), vertical distribution in the 0–5 cm and 5–10 cm layers, and horizontal distribution. Additionally, in 1982 the proportions of matures and juveniles, species composition, relative abundance of the species and the SHANNON-WIENER diversity index (H) were determined. The index is given as "e^H" in tables 6 and 7. This exponential form is closely correlated with species richness. Enchytraeid biomass was determined using the method of KITAZAWA (1977). The worms were assigned to 5 length groups <2, 2–5, 5–10, 10–15, and <15 mm. Each group was placed on a filter paper to remove excess water and then weighed on Sartorius 2434 balance.

Soil water content was measured on each sampling occasion and expressed as % of wet mass.

Statistical comparison of the means was performed using Duncan's test and the Student's t-test. The input data of enchytraeid abundance were transformed by $\sqrt{x}$. The coefficient of fidelity (V) (e.g. REISENAUER, 1965) used in Section 3.4. is a measure of the extent to which species are restricted to particular environmental conditions. Species were considered to have a "high degree of fidelity for an experimental plot" if $V > 0.3$ where $V = \frac{ad - bc}{\sqrt{(a+b)(a+c)(b+d)(c+d)}}$. The latters a, b, c and d are equivalent to those of a 2×2 contingency table.

3. Results

3.1. Enchytraeid abundance and biomass

Enchytraeid abundance in the soil of the apple orchard (table 1) ranged from 700 $ind \cdot m^{-2}$ to 23,400 $ind \cdot m^{-2}$ (averages of 5,262 to 10,678 $ind \cdot m^{-2}$). Markedly higher abundances were found in the 0–5 cm layers (averages 3,262 to 8,166 $ind \cdot m^{-2}$) than in the 5–10 cm layers (averages of 1,450 to 3,056 $ind \cdot m^{-2}$) of all the experimental plots.

Mean abundance during the year differed between plots. The highest difference was found between variant 5 (7,656 $ind \cdot m^{-2}$) and variant 40 (10,678 $ind \cdot m^{-2}$) in 1982 (table 1). The other differences recorded between the variously treated plots were not statistically significant.

The enchytraeid abundances found in 1981 were lower than those in 1982 for those plots investigated in both years. The maximum difference was found in variant 40 (5,262 $ind \cdot m^{-2}$ in 1981 and 10,678 $ind \cdot m^{-2}$ in 1982; table 1).

The seasonal dynamics of enchytraeid abundance was similar in all plots during the course of the study (fig. 1) except for the period from 3rd June to 8th July, 1981. During this time the enchytraeid density slowly decreased in the control plot from 1,800 to 1,500 to 700 $ind \cdot m^{-2}$ whereas in variants 10 and 40 the enchytraeid numbers increased; in variant 10 from 1,000 to 1,400 to 2,800 $ind \cdot m^{-2}$ and in variant 40 from 1,600 to 2,700 to 2,800 $ind \cdot m^{-2}$. The soil water content recorded during this period was: control 13.8–8.8–12.0 %, variant 10 15.8–13.5–17.0 % and variant 40 14.4–13.0–16.5 %. In 1981, a close relationship was found in the control plot between enchytraeid abundance and the water content of the soil, $r = 0.88$, $y = 1{,}274x - 13{,}541$. A similar close relationship was not found in the remaining plots.

Enchytraeid biomass in the experimental plots fluctuated between 0.37 $g \cdot m^{-2}$ and 14.00 $g \cdot m^{-2}$ (average biomass was between 3.21 and 5.36 $g \cdot m^{-2}$) (table 2). In both years the highest biomass was found in the control (3.75 $g \cdot m^{-2}$ in 1981 and 5.36 $g \cdot m^{-2}$ in 1982) (table 7). Seasonal changes in biomass resembled the changes in abundance (fig. 1). The differences between

Table 1. Enchytraeid abundance A (inds $m^{-2} \pm 1$. standard deviation) in soil of experimental plots.

variant	1981			1982	
	layer	mean A ± 1.S.D.	min–max	mean A ± 1.S.D.	min–max
control	0– 5 cm	3,262 ± 3,448	600– 8,900	4,656 ± 2,769	1,100– 9,800
	5–10 cm	2,288 ± 1,957	100– 5,600	3,056 ± 1,197	1,400– 5,300
	0–10 cm	5,500 ± 5,151	700–14,500	7,711 ± 3,648	3,000–15,100
5	0– 5 cm	–	–	4,878 ± 1,848	2,000– 8,200
	5–10 cm	not studied	–	2,778 ± 783	1,700– 3,700
	0–10 cm	–	–	7,656 ± 2,250	3,900–11,100
10	0– 5 cm	3,725 ± 6,313	800–19,200	7,722 ± 4,632	1,300–14,300
	5–10 cm	1,600 ± 1,256	0– 4,200	2,789 ± 1,356	600– 5,100
	0–10 cm	5,325 ± 7,413	1,000–23,400	10,511 ± 5,568	1,900–19,400
20	0–5 cm	–	–	5,522 ± 2,634	1,300– 8,400
	5–10 cm	not studied	–	2,689 ± 1,374	700– 4,800
	0–10 cm	–	–	8,211 ± 3,306	2,500–12,700
40	0– 5 cm	3,813 ± 3,968	800–12,400	8,166 ± 3,741	3,000–15,200
	5–10 cm	1,450 ± 651	800– 2,500	2,511 ± 1,149	600– 4,400
	0–10 cm	5,262 ± 4,492	1,600–14,900	10,678 ± 4,098	5,600–18,000

S.D. indicates seasonal variability.

Table 2. Enchytraeid biomass B (g $m^{-2} \pm 1$. standard deviation) in soil of experimental plots.

variant	1981			1982	
	layer	mean B ± 1.S.D.	min–max	mean B ± 1.S.D.	min–max
control	0– 5 cm	2.24 ± 2.17	0.18– 5.76	3.62 ± 2.34	0.47– 7.45
	5–10 cm	1.51 ± 1.81	0.01– 5.35	1.73 ± 1.14	0.95– 4.59
	0–10 cm	3.75 ± 3.95	0.37–11.11	5.36 ± 2.64	1.79– 8.56
5	0– 5 cm	–	–	2.88 ± 2.04	0.27– 7.21
	5–10 cm	not studied	–	1.98 ± 0.94	0.61– 3.34
	0–10 cm	–	–	4.86 ± 2.61	2.54–10.14
10	0– 5 cm	2.29 ± 4.04	0.25–12.24	2.78 ± 1.92	0.63– 5.38
	5–10 cm	0.92 ± 0.76	0 – 1.96	0.94 ± 0.54	0.31– 1.86
	0–10 cm	3.21 ± 4.44	0.49 ± 14.00	3.72 ± 2.19	1.12– 7.11
20	0– 5 cm	–	–	2.62 ± 1.38	0.35– 3.93
	5–10 cm	not studied	–	1.99 ± 1.29	0.69– 4.52
	0–10 cm	–	–	4.61 ± 2.01	1.73– 7.84
40	0– 5 cm	2.58 ± 2.60	0.40– 8.33	2.71 ± 1.23	0.68– 4.53
	5–10 cm	1.03 ± 0.65	0.34– 2.07	0.96 ± 0.39	0.32– 1.45
	0–10 cm	3.61 ± 2.94	0.99– 9.70	3.68 ± 1.29	1.98– 5.65

S.D. indicates seasonal variability.

enchytraeid biomasses were not statistically tested. The average biomass in variants 10 and 40 differed only slightly between years, whereas the biomass in the control in 1982 ($5.36\ g \cdot m^{-2}$) greatly exceeded that in 1981 ($3.75\ g \cdot m^{-2}$). Also, the abundances in variants 10 and 40 in 1982 were practically twice those in 1981, whereas the biomass remained the same; the average mass per individual worm in variants 10 and 40 in 1982 was approximately half of that in 1981 (table 7).

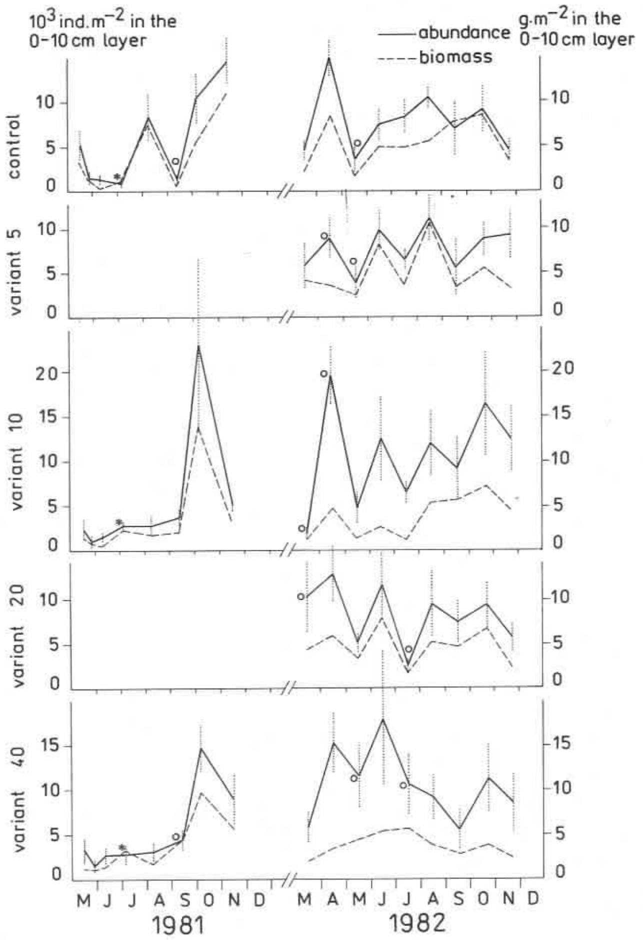

Fig. 1. Seasonal dynamics in enchytraeid abundance A ± 1 S.E. (full line) and biomass (broken line) at the control and the experimental plots treated by herbicide in doses of 5, 10, 20 and 40 kg per hectare. Statistically significant difference between the means at $P < 0.01$ ✻; at $P < 0.05$ ○; Duncan's test. S.E. (dotted) indicates standing horizontal variability.

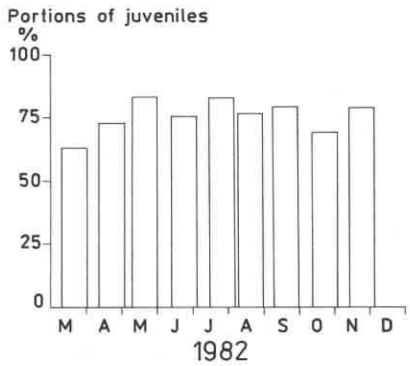

Fig. 2. Dynamics of the proportion of juvenile enchytraeids in 1982. Sum of all experimental plots.

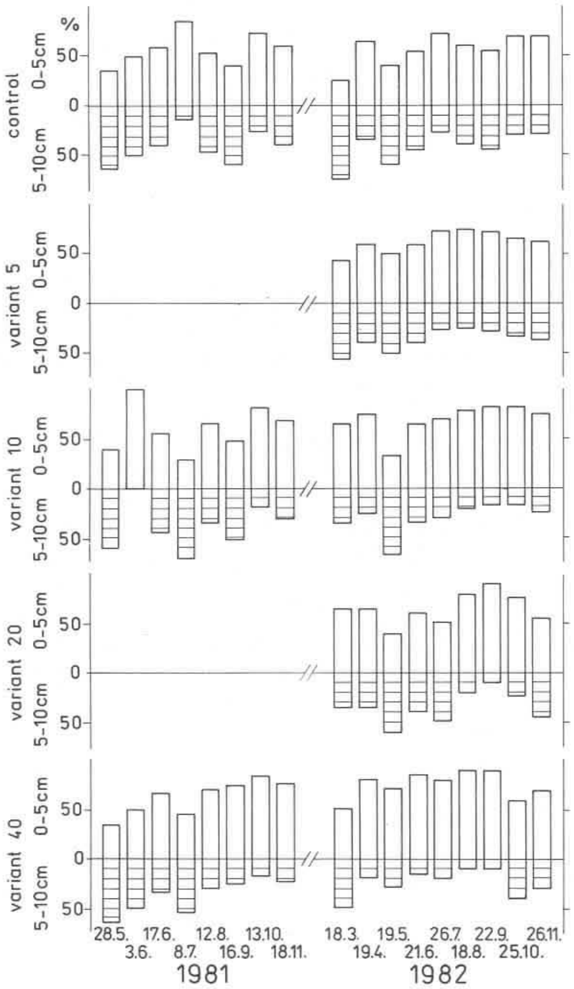

Fig. 3. Seasonal dynamics of enchytraeid vertical distribution at the control and the experimental plots treated by herbicide in doses of 5, 10, 20 and 40 kg per hectare. Percentages refer to the proportion of worms in particular layers. The 0–5 cm layer empty area, the 5–10 cm layer hatched area.

3.2. Distribution of enchytraeids in the soil

Analysis of the 1982 horizontal data showed the worms to have an aggregated distribution (S.D. $> \bar{x}$). The abundance values closely fitted a negative-binomial distribution (Chalupský & Lepš, 1985). The mean number of individuals collected in one soil core was 8.97 ± 9.68 S.D. in 1982 and 5.39 ± 7.61 in 1981.

Temporal changes in the vertical distribution of enchytraeids were studied in both years (fig. 3). Of the total number of worms, 67% and 69% inhabited the 0–5 cm layer in 1981 and 1982 respectively. The enchytraeid numbers were comparatively higher in the treated plots in this upper layer during the study.

3.3. Occurrence of mature and immature worms

Separation of adult and juveniles worms was carried out in 1982. Juveniles constituted 75.4% of the worms collected (fig. 2). Adults occurred in highest numbers towards the end of winter (18th

March, 1982), when they averaged 39.1 % of total enchytraeid abundance in the treated plots, and 73.0 % of the control population. During the rest of 1982 the proportion of adults ranged from 17.8 % to 30.1 %, and averaged 24.6 %. Fluctuations of the adult: juvenile ratio were apparent in 1982, but these were not statistically significant. Slightly higher numbers of young worms were regularly found in the chemically treated plots, compared with the control (table 7).

3.4. Species composition

In 1981, only genera were identified. *Fridericia* dominated the enchytraeid fauna in all experimental plots (D = 72.4–83.8 %) (table 3). The second most abundant genus was *Enchytraeus* (D = 15.8–19.0 %). The genera *Enchytronia* and *Henlea* were represented by a small number of individuals. Genera composition varied a little between plots. For example, there was a lower dominance of *Fridericia* (D = 72.4 %) in variant 40 compared with the control (D = 83.8 %) and variant 10 (D = 82.2 %), but variant 40 contained a higher proportion of *Enchytraeus, Enchytronia* and *Henlea*. A similar differences was observed in 1982 (table 3).

In 1982, adults were identified to species and species composition data are based on these. However, as some 75 % of individuals were juveniles, some distortion of the actual composition is likely. Unfortunately, it is only occasionally possible to identify juvenile enchytraeids to species.

The experimental plots were inhabited by a total of 17 enchytraeid species. These are listed with their dominance value in table 4. *Fridericia connata* usually had the highest dominance value. It dominated the control plot as well as variants 5, 20 and 40 (D = 3.9–11.6 %). However in variant 10 the predominant species were *F. nemoralis* (D = 5.8 %) and *F. gracilis* (D = 5.6 %).

The enchytraeid faunas of the experimental plots differed from each other through the presence of certain species which had low dominance values (table 4). Thus, *Fridericia sylvatica* was collected from variants 10 and 40 only, whereas *F. callosa* was found only in variant 5, and *Enchytronia annulata* in variants 20 and 40. *Henlea perpusilla* was not found in the control, and *H. ventriculosa* was not recorded from the control or from variant 40.

The coefficient of fidelity was calculated for each species. *Fridericia gracilis* and *Henlea ventriculosa* has a high degree of fidelity to variants 10, and 20 respectively, and *F. sylvatica* and *H. perpusilla* were particularly associated with variant 40.

The seasonal dynamics of species occurrence was studied in detail in 1982. Practically no significant differences were found between the experimental plots, thus the species densities are combined in table 5. However, I did observe a higher abundance of some adults during certain periods of the year. Adult *Fridericia connata* were frequent during April–October. Adult *F. nemoralis* occurred in spring and in autumn. Adult *F. singula, F. paranemoralis, F. bisetosa, F. alata, Enchytraeus* sp. and *E. buchholzi* tended to occur in spring, and adult *F. bulboides, F. ratzeli* and *F. gracilis* were more numerous in autumn (table 5).

The species diversity of the enchytraeid fauna was expressed by the Shannon-Wiener index. It was calculated separately for each sampling occasion and for each experimental plot (table 6). The values found for variant 5 on the 18th August and 22nd September, 1982 show how the index is influenced both by the number of species and by species frequency of occurrence. In both cases the number of species was 6. However, on the 18th August the abundance of *Fridericia connata* exceeded that of the other species almost twentyfold (index = 2.54), whereas on the 22nd September, the species were of similar abundance and the index was high (5.54).

4. Discussion

To evaluate the effect of herbicides on the soil fauna it is necessary to know how long they persist in the soil, and whether they are directly toxic to the soil fauna or act indirectly, through their effects on the vegetation and soil microflora. Triazine herbicides are very resistant to decomposition in the soil (Kearney & Kaufman, 1969; Cremlyn, 1985). Their decomposition half-life may be 1–3 years (Fuka *et al.*, 1981). Nicholls *et al.* (1982) found the half-life of atrazine was 60 d in

Table 3. Individuals of genera in experimental plots with their relative abundance [%], based on a total of matures and juveniles.

		control	variants			
			5	10	20	40
1981	*Fridericia*	83.8	–	82.2	–	72.4
	Enchytraeus	15.8	–	16.0	–	19.0
	Enchytronia	–	–	0.9	–	6.7
	Henlea	0.4	–	0.9	–	1.9
1982	*Fridericia*	85.9	86.8	79.2	79.8	71.7
	Enchytraeus	12.7	10.6	16.0	11.1	19.5
	Enchytronia	1.3	2.0	3.3	1.2	2.4
	Henlea	0.1	0.6	1.5	7.9	6.4

Table 4. Species composition in experimental plots with relative abundance [%] in 1982, based on mature worms.

species	control	variants			
		5	10	20	40
Fridericia connata BRETSCHER, 1902	9.1	11.6	1.7	8.4	3.9
F. nemoralis NURMINEN, 1970	2.3	2.8	5.8	2.2	2.5
F. singula NIELS. *et* CHRIST., 1961	3.6	2.8	1.7	1.9	1.0
F. bulboides NIELS. *et* CHRIST., 1959	0.9	1.6	1.1	1.9	2.5
F. ratzeli (EISEN, 1872)	3.5	1.3	1.8	1.6	0.5
F. gracilis VON BÜLOW, 1957	1.2	–	5.6	1.6	1.7
F. paranemoralis DÓZSA-FARKAS, 1982	2.2	1.6	0.8	1.8	0.6
F. bisetosa (LEVINEN, 1884)	1.2	1.0	1.2	0.3	0.9
F. alata NIELS. *et* CHRIST., 1959	1.6	1.0	0.2	0.5	0.3
F. sylvatica HEALY, 1979	–	–	0.3	–	1.2
F. callosa (EISEN, 1878)	–	0.6	–	–	–
Enchytraeus sp.	1.3	1.3	0.8	1.2	1.0
E. buchholzi VEJDOVSKÝ, 1878	2.0	0.1	0.8	0.4	1.2
Enchytronia sp.	0.7	0.7	1.2	0.3	0.5
E. annulata NIELS. *et* CHRIST., 1959	–	–	–	0.4	0.1
Henlea perpusilla FRIEND, 1911	–	0.3	0.3	0.3	2.0
H. ventriculosa (D'UDEKEM, 1854)	–	0.1	0.1	2.0	–

loamy-sand soil, and during the experiment the highest concentration of herbicide was in the 0–3 cm layer.

There are no unequivocal results on the toxicity of herbicides on microdrile oligochaetes in published studies. FUKA *et al.* (1981) found Zeazin not to be toxic to *Tubifex tubifex* (Tubificidae). The LC_{50} exceeded the solubility of Zeazin in water. An experiment on the impact of atrazine of *Tubifex tubifex* (AMMON, 1985), showed the herbicide in a concentration of 100 mg $\times$ l^{-1} to have a mild toxic effect after 7 d whereas 320 mg $\times$ l^{-1} was lethal in 4 d. Also the toxic influence on *Enchytraeus albidus* of Zeazin 50 in strong concentrations (0.125, 0.25, 0.50 and 1.00% water suspension) was observed in the laboratory (CHALUPSKÝ, 1984).

The impact of insecticides and nematocides on enchytraeids (MARTIN, 1975; HEUNGENS, 1968, 1969, 1972) is varied. The published information on herbicide effects on enchytraeids has been reviewed by EIJSACKERS & DRIFT (1976) and EIJSACKERS & BUND (1980). These authors concluded that enchytraeid abundance tends to be reduced by the action of herbicides. POPOVICI *et al.* (1977) reported that atrazine harmfully affected enchytraeids. LEE (1985) listed atrazine as having an evident impact on earthworms (Lumbricidae), especially in the high dose used.

In view of the present evidence that triazine herbicides may have a low toxicity to microdrile

Table 5. Number of individuals collected in all experimental plots in 1982.

	Mar 18	Apr 19	May 19	Jun 21	Jul 26	Aug 18	Sep 22	Oct 25	Nov 26	C
Fridericia connata	3	33	25	62	28	38	14	44	11	33.3
F. nemoralis	23	15	4	12	3	12	3	26	32	14.2
F. singula	7	50	3	10	3	4	1	5	1	11.3
F. bulboides	2	4	1	5	5	9	16	17	6	10.4
F. ratzeli	4	3	2	5	–	2	3	30	18	9.8
F. gracilis	9	7	11	8	3	9	16	19	7	8.9
F. paranemoralis	18	18	1	6	–	4	5	–	3	8.0
F. bisetosa	3	13	2	4	2	8	2	3	–	6.9
F. alata	6	10	–	2	1	3	–	5	–	5.1
F. sylvatica	–	–	–	3	2	2	–	7	1	2.2
F. callosa	1	–	–	–	–	–	3	–	–	0.4
Enchytraeus sp.	12	18	1	3	6	4	–	1	–	7.3
E. buchholzi	15	10	–	4	3	3	–	3	–	5.8
Enchytronia sp.	4	8	–	3	4	8	–	1	–	4.4
E. annulata	4	–	–	–	–	–	–	–	–	0.4
Henlea perpusilla	2	2	1	–	–	3	5	4	9	3.3
H. ventriculosa	–	2	–	2	2	7	3	1	–	2.0
Fridericia spp. juv.	131	355	206	325	217	334	232	305	286	83.8
Enchytraeus spp. juv.	38	125	23	118	34	67	32	49	13	40.2
Enchytronia spp. juv.	1	11	4	6	21	2	6	4	–	7.8
Henlea spp. juv.	6	23	2	6	6	–	5	28	17	8.2
Total	**289**	**709**	**286**	**584**	**340**	**519**	**346**	**552**	**404**	

C [%] – frequency of occurrence, i.e. proportion of soil cores with a species, 450 cores = 100%.

oligochaetes (here Enchytraeidae and Tubificidae), we assume that Zeazin 50 in our study did not have a decisively toxic impact on the enchytraeids. Therefore, the observed changes in enchytraeid abundance and number of species probably reflected indirect effects of the use of the herbicide.

After the application of Zeazin 50 the grass understorey was depleted in the experimental variants 5, 10, 20 and 40. The dead plant matter remained on the surface available to soil organisms. Killing the grass must have changed the rate of evapotranspiration of water from the soil, but the periodic measurements of soil water content gave a poor indication of these changes (table 7). The maxima and minima of soil moisture were missed.

The experimental sites were not investigated before the herbicide application on 28th May, 1981. The influence of Zeazin 50 can therefore be determined only by comparing the treated sites with the control. The homogeneity of the plots was tested on the first sampling occasion, shortly before the treatment. The differences between the enchytraeid abundances in the plots were not statistically significant at this time (fig. 1, compare variants 10 and 40 and the control).

Statistical treatment demonstrated significant differences between the enchytraeid abundances of the sites in 8 cases (Section 3.1., Fig. 1). In all these cases significantly higher abundance was observed at those sites which were treated with the higher dose of Zeazin 50. This finding is contrary to the expected effect of the herbicide. The enchytraeid densities were likely to have been affected by factors other than the herbicide alone. Slightly higher soil water content in some plots and increased food supply from dead plants may have resulted in increased worm abundance, although this suggestion is only weakly supported by the soil moisture data.

Enchytraeid abundance increased in variants 10 and 40 in 1982. Also the proportion of juveniles was higher in these plots although at the same time the total population biomass decreased. A possible explanation is that conditions appeared in the treated plots which were favourable for higher reproductive activity producing an increase in total enchytraeid abundance and in the proportion of young worms. Perhaps this situation was caused by a greater number of smaller

Table 6. Shannon-Wiener index of species diversity (e^H) and number of species (in brackets) for each sampling occasion and experimental plot in 1982.

variant	Mar 18	Apr 19	May 19	Jun 21	Jul 26	Aug 18	Sep 22	Oct 25	Nov 26	mean ±1.S.D.
control	8.04 (10)	7.62 (9)	2.00 (2)	3.03 (7)	4.34 (7)	5.66 (7)	4.59 (6)	4.87 (9)	3.71 (4)	4.87 ± 1.99 (6.8 ± 2.5)
5	6.64 (9)	6.17 (9)	3.17 (4)	2.79 (6)	4.80 (6)	**2.54** (6)	**5.45** (6)	2.81 (6)	5.43 (7)	4.42 ± 1.60 (6.6 ± 1.6)
10	4.46 (5)	10.46 (13)	2.42 (4)	6.93 (8)	5.35 (6)	9.15 (11)	3.05 (6)	5.11 (9)	3.33 (6)	5.58 ± 2.77 (7.6 ± 3.0)
20	8.49 (12)	7.74 (10)	1.66 (3)	3.88 (7)	1.00 (1)	3.56 (5)	4.70 (6)	4.93 (7)	3.92 (5)	4.43 ± 2.46 (6.2 ± 3.3)
40	6.70 (10)	8.58 (10)	4.77 (6)	9.60 (12)	4.51 (7)	7.28 (9)	2.87 (3)	7.20 (9)	4.05 (5)	6.17 ± 2.24 (7.9 ± 2.8)

Table 7. Fluctuations of determined characteristics between the experimental plots (c – the control).

characteristic	1981						1982									
	variant			data			variant					data				
mean water content in soil [%] ± 1. S.D.	c	40	10	15.0 ± 3.5	16.4 ± 1.8	16.7 ± 1.9	5	c	10	20	40	13.5 ± 4.0	13.7 ± 2.4	14.0 ± 3.0	14.8 ± 2.1	14.9 ± 3.0
mean enchytraeid abundance (individuals per sq.m)	40	10	c	5,262	5,325	5,550	5	c	20	10	40	7,656	7,711	8,211	10,511	10,678
mean enchytraeid biomass (g per sq.m)	c	40	10	3.75	3.61	3.21	c	5	20	10	40	5.36	4.86	4.61	3.72	3.67
mean mass of one individual [mg]	40	c	10	0.69	0.68	0.60	c	5	20	10	40	0.69	0.64	0.56	0.35	0.34
proportion of juveniles in population [%]							c	5	20	10	40	71	73	75	77	80
proportion of worms in the 0–5 layer (% of total 0–10 cm)	c	10	40	59	70	72	c	5	20	10	40	60	63	67	73	76
mean Shannon-Wiener's index species diversity (e^H)							5	20	c	10	40	4.42	4.43	4.87	5.58	6.17
mean number of species at sampling occasion							20	5	c	10	40	6.2	6.6	6.8	7.6	7.9
total number of species at experimental plot							c	5	10	20	40	12	14	15	15	15

species with shorter life cycles. As a result, the plots were inhabited by a more abundant enchytraeid population with a lower total biomass. This is reminiscent of the general theory about changes which happen in an animal community after a temporary enhancement of food sources and their fast utilization.

Analysis of the enchytraeid community at the species and genus levels indicated some differences between the plots. These differences concerned the dominance of species and genera of rarely occurring species at some sites (Section 3.4.). These differences were so small that they could hardly be connected with the application of herbicide alone. Rather, they reflected the whole complex of abiotic and biotic changes associated with the herbicide treatment. The observed changes in dominance and frequency of enchytraeids occurrence support the opinion of EIJSACKERS & BUND (1980). These authors pointed (p. 292–293) to the fact that during field studies on the effect of herbicides on soil fauna only minute changes in the species composition of soil community might happen. These changes usually involved only infrequent occurring species and it is difficult to interpret them as they may be related to many other circumstances, not only to the presence of a herbicide.

In conclusion, differences between the enchytraeid faunas of the experimental plots, following the application of Zeazin 50, were either within the natural range of seasonal variability or could have been caused by the slight non-homogeneity within the study site at the beginning of the experiment. Moreover, the enchytraeid may have been slightly affected by the parallel changes in soil water content and by the increase in food supply from dead plants. There was no evidence that single doses of Zeazin 50 up to 40 kg per hectare adversely affected the enchytraeid fauna of the apple orchard soil studied.

5. Zusammenfassung

[Der Einfluß von Zeazin 50 auf die Enchytraeidae (Oligochaeta) im Boden eines Apfelgartens]

Herbizide sind die am meisten in der Landwirtschaft benutzten Pestizide. In den Jahren 1981–1982 wurde der Einfluß des Herbizides Zeazin 50 in den Dosen von 0, 5, 10, 20 und 40 $kg \cdot ha^{-1}$ auf die Enchytraeidae in einem Feldversuch in Südböhmen untersucht. Dabei wurden 17 Enchytraeiden-Arten festgestellt. Die aktuelle Abundanz der Enchytraeiden variierte von 700 bis 23,400 $ind \cdot m^{-2}$, die mittlere Abundanz von 5,262 bis 10,678 $ind \cdot m^{-2}$. Die aktuelle Biomasse schwankte zwischen 0,37 und 14,00 $g \cdot m^{-2}$, die mittlere Biomasse zwischen 3,21 und 5,36 $g \cdot m^{-2}$. Es wurden nur geringe Veränderungen in der Enchytraeiden-Population festgestellt. Im Vergleich zur Kontrolle war der Enchytraeiden-Besatz im Versuch mit 40 kg Zeazin $\cdot ha^{-1}$ leicht erhöht. Wegen des höheren Anteils an juvenilen Tieren war die Biomasse geringer, doch die Zahl der Arten war größer, und diese Differenzen wurden wahrscheinlich, eher indirekt (durch Veränderungen der Bodenfeuchtigkeit, des Gehaltes an pflanzlicher Nekromasse sowie durch den ganzen Komplex von biotischen und abiotischen Faktoren) als direkt durch Wirkung des Herbizides verursacht.

Schlüsselwörter: Enchytraeidae, Braunerde, Herbizide, Saisondynamik, Artenbestand.

6. Acknowledgement

The expert discussion of Dr. JOSEF RUSEK CSc. and Dr. MARTA TESAŘOVÁ CSc. is gratefully acknowledged. I also want to thank Dr. MICHAEL A. LEARNER, who rectified the English of this manuscript and added some helpful suggestions. Dr. MARTIN TONNER translated the summary into German.

7. Literature

ABRAHAMSEN, G., 1969. Sampling design in studies of population densities in Enchytraeidae (Oligochaeta). Oikos **20**, 54–66.

AMMON, U. H., 1985. Worm toxicity tests using *Tubifex tubifex*. *In:* Anonymous, Compartement et effets secondaires des pesticides dans le sol. Colloq. I.N.R.A., **31**, 303–317.

CHALUPSKÝ, J., 1984. [To the ecology and taxonomy of enchytraeids (Enchytraeidae) of fruit growing region by Vodňany in South Bohemia]. Ph. D. Thesis, Karlova universita, Praha, 221 pp. (In Czech.).

– J. LEPŠ, 1985. The spatial pattern of Enchytraeidae (Oligochaeta). Oecologia (Berlin), **68**, 153–157.

CREMLYN, R., 1985. [Pesticides – Preparation and Mode of Action]. SNTL, Praha, 244 pp. (Czech. translation).

EIJSACKERS, H., & C. F. VAN DE BUND, 1980. Effects on soil fauna. *In:* R. J. HANCE (ed.), Interactions between herbicides and the soil. Academic Press, London, New York, Toronto, 255–305.
– J. VAN DER DRIFT, 1976. Effects on the soil fauna. *In:* L. J. AUDUS (ed.), Herbicides. Physiology, Biochemistry, Ecology, vol. II. Academic Press, London, New York, San Francisco, 149–174.
FUKA, T., J. HAVLÍKOVÁ, Z. SVOBODOVÁ, J. ŠAUER, J. KOPŘIVA, 1981. [Biological decomposability and toxicity of selected pesticides to aquatic organisms]. *In:* Anonymous, Spoločenský význam zoologických výskumov pri tvorbe a ochrane životného prostredia. Bratislava, 66–73 (In Czech.).
HEUNGENS, A., 1968. The influence of DBCP on the soil fauna in azalea culture. Pedobiologia **8**, 281–288.
– 1969. L'influence de la fumure et des pesticides aldrine, carbaryl et DBCP sur la faune du sol dans la culture des Azalées. Rev. Écol. Biol. Sol. **6**, 131–145.
– 1972. Involved van hygromull op de bodemfauna in de azaleateelt. Meded. Fac. Landb.-Wet., Rijksuniv. Gent, **37**, 824–830.
KEARNEY, P. C., & D. D. KAUFMAN, 1969. Degradation of Herbicides. Marcel Dekker Inc., New York, 394 pp.
KITAZAWA, T., 1977. Numbers and biomass of Enchytraeidae. *In:* Y. KITAZAWA (ed.), Ecosystem analysis of the subalpine coniferous forest of the Shigayama IBP area, central Japan (JIBP Synthesis 15), 110–114.
LEE, K. E., 1985. Earthworms. Their ecology and relationships with soils and land use. Academic Press, Sydney, Tokyo, 411 pp.
MARTIN, N. A., 1975. Effect of four insecticides on the pasture ecosystem. IV. Enchytraeidae and Diptera larvae heat-extracted in water-filled funnels. New Zealand J. Agric. Res. **18**, 313–315.
NICHOLLS, P. H., A. WALKER, & R J. BAKER, 1982. Measurement and simulation of the movement and degradation of Atrazine and Metribuzin in a fallow soil. Pestic. Sci. **12**, 484–494.
O'CONNOR, F. B., 1955. Extraction of Enchytraeid worms from a coniferous forest soil. Nature (London) **175**, 815–816.
POPOVICI, I., G. STAN, V. STEFAN, R. TOMESCU, A. DUMEA, A. TARTA, & F. DAN, 1977. The influence of Atrazine on soil fauna. Pedobiologia **17**, 209–215.
REISENAUER, R., 1965. Metody matematické statistiky a jejich aplikace. SNTL – Práce, Praha, 210 pp.

Synopsis: *Original scientific paper*

CHALUPSKÝ, J., 1989. Influence of Zeazin 50 on Enchytraeidae (Oligochaeta) in an apple orchard soil. Pedobiologia **33**, 361–371.

In South Bohemia, Czechoslovakia, the influence on Enchytraeidae of the herbicide Zeazin 50, applied in doses of 0, 5, 10, 20 and 40 kg per hectare, was analysed in a field experiment during 1981–1982. Seventeen enchytraeid species were recorded in this study. Enchytraeid abundances on individual sampling dates ranged from 700 to 23,400 ind $\cdot$ m^{-2}, and average abundance per treatment from 5,262 to 10,678 ind $\cdot$ m^{-2}. Seasonal biomass fluctuated between 0.37 and 14.00 g $\cdot$ m^{-2}, and mean biomass per treatment between 3.21 and 5.36 g $\cdot$ m^{-2}. Only minute changes in enchytraeid populations were observed. In comparison with the control, the enchytraeid in the plot treated with 40 kg of Zeazin 50 per hectare tended to occur in slightly higher numbers bur with a lower biomass. The population also contained a higher proportion of juveniles, and a higher number of species. These differences were probably caused by changes in soil water content and in dead plant matter supply, and by the whole complex of abiotic and biotic conditions associated with the application of the herbicide, rather than by the herbicide itself.
Key words: Enchytraeidae, brown soil, herbicides, seasonal dynamics, species community.

Address of the author: Dr. J. CHALUPSKÝ, Ústav půdní biologie ČSAV, Na sádkách 7, CS - 37005 České Budějovice (Czechoslovakia)